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ORIGINAL ARTICLE

Retinal nerve fiber layer and ophthalmic artery blood flow parameters in pseudoexfoliation syndrome and pseudoexfoliative glaucoma

Paramètres de la couche des fibres nerveuses rétiniennes et du débit sanguin de l'artère ophtalmique dans le syndrome de pseudo-exfoliation et le glaucome pseudo-exfoliatif

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KEYWORDS
Color Doppler ;
Glaucoma ;
Pseudoexfoliation ;
Ophthalmic artery ;
Retinal nerve fiber layer

Summary
Objectives. — To investigate the relationship between ophthalmic artery (OA) blood flow parameters and retinal nerve fiber layer (RNFL) thickness in eyes with pseudoexfoliation (PEX).
Methods. — We compared PEX eyes without glaucoma (group A, $n = 53$) and those with glaucoma (group B, $n = 18$) with control eyes (group C, $n = 44$). Subsequently, eyes in groups A and B were compared. Finally, OA color Doppler imaging measurements were recorded, and peripapillary RNFL analysis was performed.
Results. — The total RNFL measurements differed significantly among the groups ($P = 0.012$), being thicker in group C than in group A ($P = 0.010$) and significantly different between group B and groups A and C (both $P = 0.001$). The peak systolic velocity (PSV) and end diastolic velocity (EDV) measurements of groups A and B were lower than those of group C (PSV: $P = 0.001$ and $P = 0.001$, respectively; EDV: $P = 0.001$ and $P = 0.001$, respectively). No significant difference was noted in resistive index (RI) measurements ($P = 0.370$). In group B, significant negative correlations were noted between total RNFL and PSV ($r = -0.743$; $P = 0.001$) and between total RNFL and EDV ($r = -0.691$; $P = 0.001$), but not between total RNFL and RI measurements ($P = 0.548$).

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Conclusions. — Pseudoexfoliation syndrome (PXS) with or without glaucoma was associated with a decrease in the PSV and EDV values of the OA. An extensive study may be needed to further explore the role of PXS in OA blood flow parameters. Total RNFL thickness values were lower in eyes with PEX than in those without.

MOTS CLÉS
Doppler Couleur ;
Glaucome ;
Pseudo-exfoliation ;
Artère ophtalmique ;
Couche fibreuse du nerf rétinien

Résumé
Objectifs. — Étudier le lien entre les paramètres du débit sanguin de l'artère ophtalmique (AO) et l'épaisseur de la couche des fibres nerveuses rétiniennes (CFNR) dans les yeux présentant une pseudo-exfoliation (PEX).
Méthodes. — Nous avons comparé des yeux PEX sans glaucome (groupe A, $n = 53$) et avec glaucome (groupe B, $n = 18$) avec des yeux témoins (groupe C, $n = 44$). Par la suite, les yeux des groupes A et B ont été comparés entre eux. Enfin, des relevés d'imagerie Doppler Couleur de l'AO et une analyse de la CFNR péripapillaire ont été effectués.
Résultats. — Les relevés de la CFNR totale présentaient des différences significatives selon les groupes ($p = 0{,}001$). Ils étaient plus élevés dans le groupe C que dans le groupe A ($p = 0{,}010$) et présentaient des différences significatives entre les groupes B et C ($p = 0{,}001$). Les relevés de la vitesse maximale systolique (VMS) et de la vitesse diastolique finale (VDF) dans les groupes A et B étaient inférieurs à ceux du groupe C (respectivement $p = 0{,}007$ et $p = 0{,}001$; respectivement $p = 0{,}004$ et $p = 0{,}001$).
Conclusions. — Le syndrome de pseudo-exfoliation (SPE) avec ou sans glaucome a été associé à une diminution des valeurs VMS et VDF de l'AO. Une étude plus approfondie peut être nécessaire pour explorer davantage le rôle du SPE dans les paramètres du débit sanguin de l'AO. Les valeurs de l'épaisseur totale de la CFNR étaient réduites dans les yeux présentant une PEX.

Introduction

Pseudoexfoliation syndrome (PXS) is an age-related syndrome. It is characterized by the production and progressive storage of small, white deposits of fibrillary, extracellular, pseudoexfoliative material in ocular tissues [1]. Pseudoexfoliative material can be observed in ocular and extraocular tissues such as those of the lens capsule, ciliary body, anterior and posterior surface of the iris, pupillary rim, trabecular meshwork, conjunctival stroma, corneal endothelium, extraocular muscles, optic nerve meninges, orbital connective components, skin, heart, kidney, bladder, lungs, liver, cerebral meninges, and adventitia of the aorta and cerebral artery [2—4].

Blood flow parameters can be altered by storing pseudoexfoliative material in the walls of iris veins, posterior ciliary arteries, vortex veins, and central retinal veins. This can lead to increased permeability, narrowing, and eventually obstruction [1]. In a study conducted in patients with at least one transient ischemic attack, the prevalence of pseudoexfoliative material was high among those with high resistive indices (RIs) for ophthalmic arteries (OAs) [5].

Color Doppler imaging (CDI) is a reliable, non-invasive, and reproducible method for the evaluation of orbital blood flow velocities. It has been increasingly used to assess pathologies such as vascular occlusion, tumors, glaucoma, and caroticocavernous fistula that affect orbital hemodynamics [6—9].

Visual field evaluation and optical coherence tomography (OCT) can provide functional data and information on the anatomical structure of the retinal nerve fiber layer (RNFL) [10]. OCT studies have primarily revealed early structural changes in the thickness of the RNFL in patients with PXS [11—13]. Some studies suggest that OCT is more sensitive and practical than other tests (e.g., visual field testing) for the detection of glaucomatous damage in the optic nerve head [14—16]. The aim of this study was to evaluate the relationship between OA blood flow parameters and RNFL thickness in eyes with pseudoexfoliation (PEX).

Materials and methods

This prospective study was performed in accordance with the principles of the Declaration of Helsinki. The study was reviewed and approved by the Ethics Committee of our university (details blinded for peer review). All patients provided written informed consent.

All patients underwent routine ophthalmologic examination. The best-corrected visual acuities were obtained, slit-lamp biomicroscopic examinations were performed before and after dilating the pupil, intraocular pressure

(IOP) was measured, angle examinations were performed with a Goldmann three-mirror lens, fundus examinations were performed with a 90-D lens, and the RNFL was assessed via OCT. RNFL data were analyzed, and the total retinal thickness and four-quadrant nerve and fiber layer values were compared. Detailed medical histories were obtained from all patients. Patients with cardiovascular disease other than systemic hypertension and patients with diabetic retinopathy were excluded from the study. Patients with systemic hypertension that was under control with medication were included in the study.

Patients with pseudoexfoliative material on the pupillary rim or anterior capsule surface after mydriasis were included in the group of patients with eyes with PEX. Patients with an IOP < 21 mmHg, diurnal IOP fluctuation within normal limits ($\leq$ 5 mmHg), no glaucomatous optic nerve changes, and normal visual field scan results were included in the group of patients with PXS. Patients with pseudoexfoliative material and glaucomatous optic nerve changes (vertical linear cup-to-disc ratio > 0.6, notching of the neuroretinal rim) upon pathological visual field tests were included in the group of patients with pseudoexfoliative glaucoma. We included patients without PEX, glaucoma, diabetic retinopathy, uncontrolled systemic hypertension, and cardiovascular pathology in the control group. We confirmed the absence of PEX in the patients in the control group while their pupils were dilated. The patients in our study were divided into three groups: patients with PEX and without glaucoma were included in group A, patients with PEX and glaucoma in group B, and control patients in group C. Three comparisons were made in the study. First, eyes of patients in the control group were compared with eyes of patients with PEX without glaucoma. Second, eyes of patients with PEX and glaucoma were compared with eyes of patients with PEX without glaucoma. Lastly, eyes of those with glaucoma and eyes of those in the control group were compared.

OA CDI was performed in all patients by the same experienced radiologist (U.I.), using Doppler sonography (Affinity 70G, probe: 5—12 MHz; Philips Healthcare, Best, the Netherlands). The radiologist performed the imaging while being blinded to group allocation. CDI was performed using a 7.5-MHz linear transducer. All imaging examinations were performed with patients in the supine position and eyes closed. Acoustic gel was applied to the eyelids of the patients, and no pressure was applied to the globe during imaging. Patients were asked to look straight ahead. First, an image of the optic nerve was obtained. The OA is situated either above or below the optic nerve in the posterior orbit before passing forward to the nasal orbit in a horizontal plane slightly superior to the optic nerve [6]. Additionally, OA blood parameters were obtained approximately 25 mm behind the globe, where the artery crossed the optic nerve [7].

Peripapillary RNFL analysis was performed with a spectral-domain OCT (3D OCT-1 Maestro; Topcon, Tokyo, Japan) device. Each papilla was analyzed at four quadrants: temporal quadrant thickness (316°—45°), superior quadrant thickness (46°—135°), nasal quadrant thickness (136°—225°), and inferior quadrant thickness (226°—315°). In addition, the total RNFL thickness was evaluated for each eye included in the study.

Statistical analysis

NCSS 2007 software (NCSS, Kaysville, UT, USA) was used for statistical analysis. Descriptive statistical methods (mean, standard deviation, frequency, ratio, minimum, and maximum) were used to evaluate the study data. The distributions of quantitative data were tested for normality using the Kolmogorov—Smirnov test, Shapiro Wilk's test, and graphical evaluations. One-way analysis of variance (ANOVA) was used to compare three or more normally distributed groups, and the Bonferroni test was used for paired comparisons. Significance was assigned at the $P < 0.05$ level.

Results

The study was conducted between March 2020 and March 2021, with 115 eye measurements recorded for 95 patients. Eyes of patients with PEX were included in groups A and B, with 14 (27.5%) cases of the right eye only, 17 (33.3%) of the left eye only, and 20 (39.2%) of both eyes. Glaucoma was not detected in 55 (74.6%; 53 patients) eyes and was observed in 18 (25.4%) (Table 1).

A significant difference was noted in peak systolic velocity (PSV) among the three groups, as determined by one-way ANOVA ($F = 8.492$; $P = 0.001$). No significant difference was noted between group A (41.26 ± 15.45 cm/s) and group B (39.54 ± 15.41 cm/s) in terms of PSV (mean difference [A-B] = 1.717 ± 4.20 cm/s; $P > 0.99$). The PSV in group A was significantly lower than that in group C (52.94 ± 15.34 cm/s; mean difference [A—C] = -11.68 ± 3.14 cm/s; $P = 0.001$). Furthermore, the PSV in group B was significantly lower than that in group C (mean difference [B—C] = -13.39 $\pm$ 4.30 cm/s, $P = 0.001$) (Fig. 1).

A significant difference was noted in end diastolic velocity (EDV) among the three groups, as determined by one-way ANOVA ($F = 8.604$; $P = 0.001$). No significant difference was noted between group A (12.45 ± 5.71 cm/s) and group B (11.54 ± 5.68 cm/s) in terms of EDV (mean difference [A—B] = 0.91 ± 1.50 cm/s, $P > 0.99$). The EDV in group A was significantly lower than that in group C (16.56 ± 5.19 cm/s; mean difference [A—C] = -4.107 $\pm$ 1.12 cm/s; $P = 0.001$). Additionally, the EDV in group B was significantly lower than that in group C (mean difference [B—C] = -5.025 ± 1.54 cm/s; $P = 0.001$) (Fig. 2).

No significant differences were noted in the RI measurements among the three groups, as determined by one-way ANOVA ($F = 1.004$; $P = 0.370$).

A significant difference was observed in total RNFL thickness among the three groups, as determined by one-way ANOVA ($F = 4.590$; $P = 0.012$). The total RNFL in group A (87.91 ± 14.97 μm) were significantly thinner than that in group C (96.18 ± 11.90 μm; mean difference [A—C] = -8.276 ± 2.75 μm; $P = 0.010$). The difference in total RNFL thickness between groups A and B (72.33 ± 2.35 μm) was significant (mean difference [A—B] = 15.58 ± 12.62 μm; $P = 0.001$). The difference (mean difference [B—C] = -23.85 ± 9.55 μm; $P = 0.001$) between groups B and C was significant (Tables 2 and 3 and Fig. 3).

A significant difference was noted in RNFL superior quadrant (RNFL S) thickness among the three groups, as

Table 1 Demographic features.

		Total ($n = 95$)	[a]PEX (+) Glaucoma (−) ($n = 37$)	[b]PEX (+) Glaucoma (+) ($n = 14$)	[c]Control ($n = 44$)
Age (years)	Mean ± SD	70.4 ± 8.93	73.81 ± 7.46	76.57 ± 9.21	65.86 ± 7.65
Sex; *n* (%)	Female	53 (55.8)	22 (59.5)	6 (42.9)	25 (56.8)
	Male	42 (44.2)	15 (40.5)	8 (57.1)	19 (43.2)

PEX: pseudoexfoliation.
[a] PEX (+) Glaucoma (−) group.
[b] PEX (+) Glaucoma (+) group.
[c] Control group.

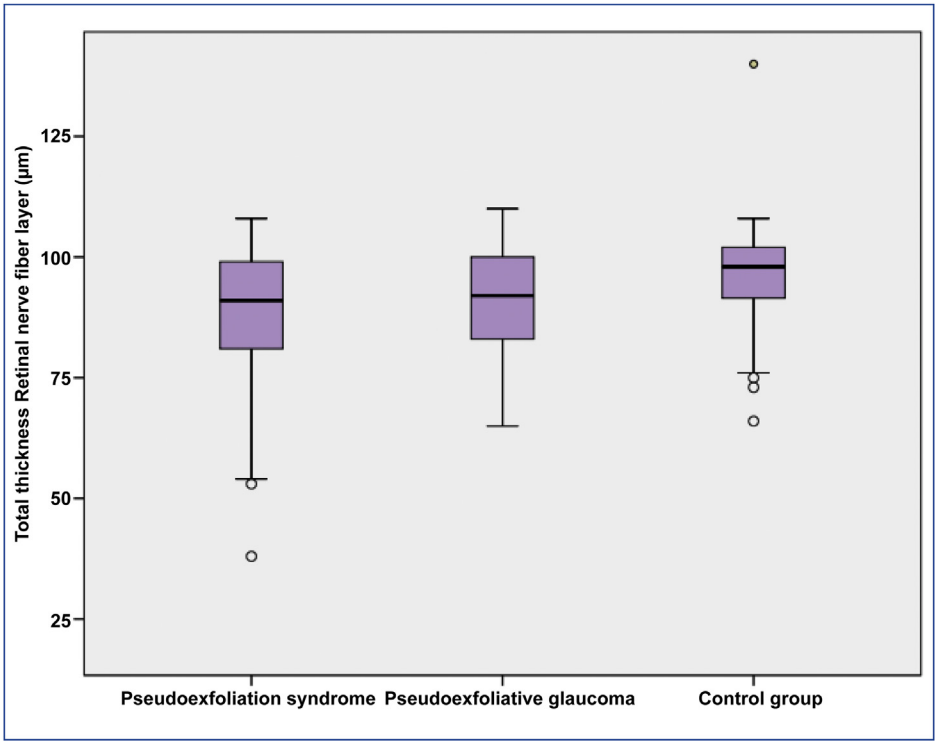

Figure 1. Peak systolic velocity distribution by group.

determined by one-way ANOVA (F = 15.002; $P = 0.001$). That of group A (100.51 ± 23.12 µm) was significantly thicker than that of group B (83.11 ± 23.80 µm; mean difference [A-B] = 17.398 ± 5.80 µm; $P = 0.010$) and significantly thinner than that of group C (114.80 ± 17.53 µm; mean difference [A-C] = −14.286 ± 4.3 µm; $P = 0.004$). RNFL S of group B was significantly thinner than that of group C (mean difference [B−C] = −31.68 ± 5.94 µm; $P = 0.001$) (Fig. 4).

A significant difference was noted in RNFL inferior quadrant (RNFL I) thickness among the three groups, as determined by one-way ANOVA (F = 12.949; $P = 0.001$). That of group A (109.87 ± 20.63 µm) was significantly thicker than that of group B (84.44 ± 27.82 µm; mean difference [A−B] = 25.423 ± 6.25 µm; $P = 0.001$; $P < 0.01$) but not significantly different from that of group C (116.91 ± 23.44 µm; mean difference [A−C] = −7.041 ± 4.67 µm; $P = 0.405$). Moreover, RNFL I of group B was significantly thinner than that of group C (mean difference [B−C] = −32.46 ± 6.42 µm; $P = 0.001$) (Fig. 4).

A significant difference was noted in RNFL temporal quadrant (RNFL T) thickness among the three groups, as determined by one-way ANOVA (F = 7.971; $P = 0.001$). No significant difference was noted between group A (67.30 ± 13.61 µm) and group B (59.94 ± 18.22 µm) in terms of RNFL T thickness (mean difference [A-B] = 7.35 ± 3.84 µm; $P = 0.174$). That of group A was significantly thinner than that of group C (74.91 ± 12.73 µm; mean difference [A−C] = −7.607 ± 2.87 µm; $P = 0.028$). In addition, the RNFL T in group B was significantly thinner than that in group C (mean difference [B−C] = -14.96 ± 3.94 µm; $P = 0.001$) (Fig. 5).

No significant difference was noted in RNFL nasal quadrant (RNFL N) thickness among the three groups, as determined by one-way ANOVA (F = 0.393; $P = 0.676$; $P > 0.05$).

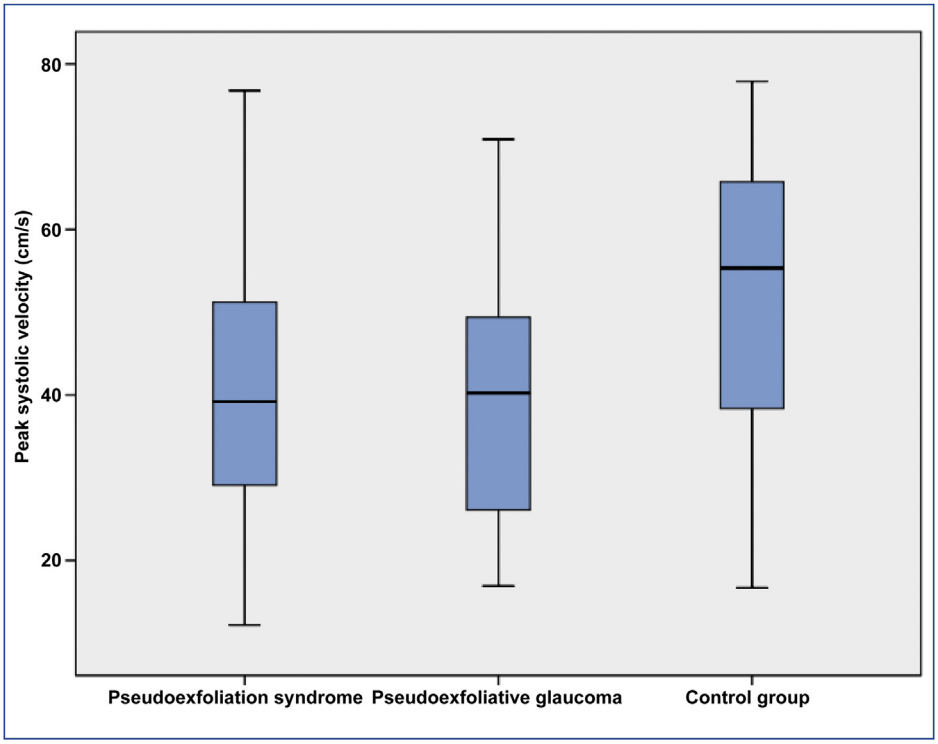

Figure 2. End diastolic velocity distribution by group.

Table 2 Total RNFL, PSV, EDV, and RI measurements..

	[a]PEX (+) Glaucoma (—) ($n=53$) Mean ± SD	[b]PEX (+) Glaucoma (+) ($n=18$) Mean ± SD	[c]Control ($n=44$) Mean ± SD	P[d]	Post Hoc P_{1-2}[e]	P_{1-3}[e]	P_{2-3}[e]	
Total RNFL thickness (μm)	87.91 ± 14.97	72.33 ± 2.35	96.18 ± 11.90	0.012*	0.001	0.010*	0.001	3 > 1
PSV (cm/s)	41.26 ± 15.45	39.54 ± 15.41	52.94 ± 15.34	0.001**	> 0.99	0.001**	0.001**	3 > 1, 2
EDV (cm/)	12.45 ± 5.71	11.54 ± 5.68	16.56 ± 5.19	0.001**	> 0.99	0.001**	0.001**	3 > 1, 2
RI	0.70 ± 0.06	0.70 ± 0.07	0.68 ± 0.06	0.370	> 0.99	0.765	0.672	—

PEX: pseudoexfoliation; RNFL: retinal nerve fiber layer; PSV: peak systolic velocity; EDV: end diastolic velocity; RI: resistive index.
* $P < 0.05$.
** $P < 0.01$.
[a] PEX (+) Glaucoma (—) group.
[b] PEX (+) Glaucoma (+) group.
[c] Control group.
[d] One-way ANOVA.
[e] Bonferroni test.

A weak negative correlation was noted between total RNFL and PSV and EDV measurements in group A, but this correlation was not significant ($P > 0.05$). No significant relationship was noted between total RNFL and RI measurements, either ($P > 0.05$).

In group B, a significant negative correlation was noted between total RNFL and PSV (r = — 0.743; $P = 0.001$). In addition, a significant negative correlation was noted between total RNFL and EDV (r = — 0.691; $P = 0.001$), whereas no significant relationship was noted between total RNFL and RI ($P = 0.548$).

In group C, a moderately significant positive correlation was noted between total RNFL and PSV (r = 0.317; $P = 0.036$) and between total RNFL and EDV (r = 0.337 $P = 0.025$). No significant relationship was noted between total RNFL and RI ($P = 0.0766$) (Table 4).

Table 3 Evaluation of RNFL measurements.

	[a]PEX (+) Glaucoma (−) ($n=53$) Mean ± SD	[b]PEX (+) Glaucoma (+) ($n=18$) Mean ± SD	[c]Control ($n=44$) Mean ± SD	*Pa*[d]	Post Hoc P_{1-2}[e]	P_{1-3}[e]	P_{2-3}[e]	
RNFL S (μm)	100.51 ± 23.12	83.11 ± 23.80	114.80 ± 17.53	0.001**	0.010*	0.004**	0.001**	3 > 1 > 2
RNFL I (μm)	109.87 ± 20.63	84.44 ± 27.82	116.91 ± 23.44	0.001**	0.001**	0.405	0.001**	1, 3 > 2
RNFL T (μm)	67.30 ± 13.61	59.94 ± 18.22	74.91 ± 12.73	0.001**	0.174	0.028*	0.001**	3 > 1, 2
RNFL N (μm)	72.53 ± 17.70	66.39 ± 23.13	74.05 ± 17.70	0.676	0.605	> 0.99	0.435	—
Total RNFL (μm)	87.91 ± 14.97	72.33 ± 2.35	96.18 ± 11.90	0.001**	0.001**	0.044*	0.001**	1,3 > 2

PEX: pseudoexfoliation; RNFL S: retinal nerve fiber layer superior quadrant; RNFL I: retinal nerve fiber layer inferior quadrant; RNFL T: retinal nerve fiber layer temporal quadrant; RNFL N: retinal nerve fiber layer nasal quadrant.
* $P < 0.05$.
** $P < 0.01$.
[a] PEX (+) Glaucoma (−) group.
[b] PEX (+) Glaucoma (+) group.
[c] Control group.
[d] One-way ANOVA.
[e] Bonferroni test.

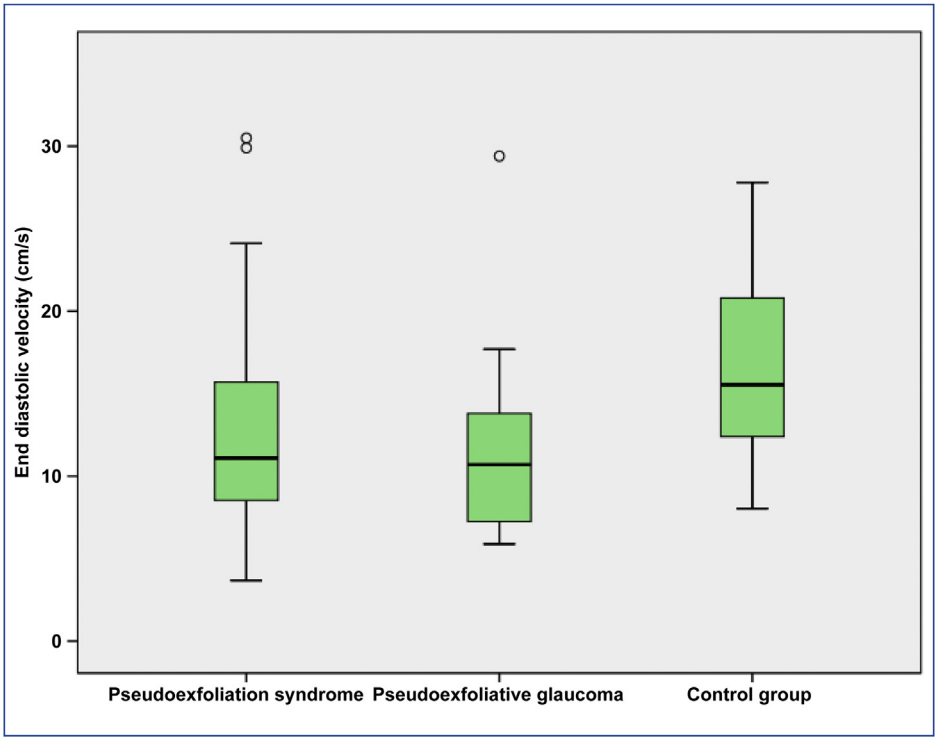

Figure 3. Total parameters and retinal nerve fiber layer thickness distribution by group.

Discussion

This study showed that patients with PEX with and without glaucoma had differences in the hemodynamic parameters of their OAs and thinner RNFLs. Aging is associated with decreased EDV values for unchanged PSV values of the OA [17]. In a previous study [18], changes in blood flow velocity resulted in an increased vascular RI; therefore, an increased RI in the analyzed group was associated with an increased systolic blood pressure (SBP), while a decreased RI was associated with an increased diastolic blood pressure (DBP). In addition, an increased DBP correlates with an increased EDV, which leads to a decreased RI. Similarly, an increased SBP and PSV cause an increased RI in the OA. These observations confirm the age-related vascular remodeling theory. Structural changes decrease the diameter of the vessels and increase their rigidity. This remodeling provides the stable velocity of the current in the

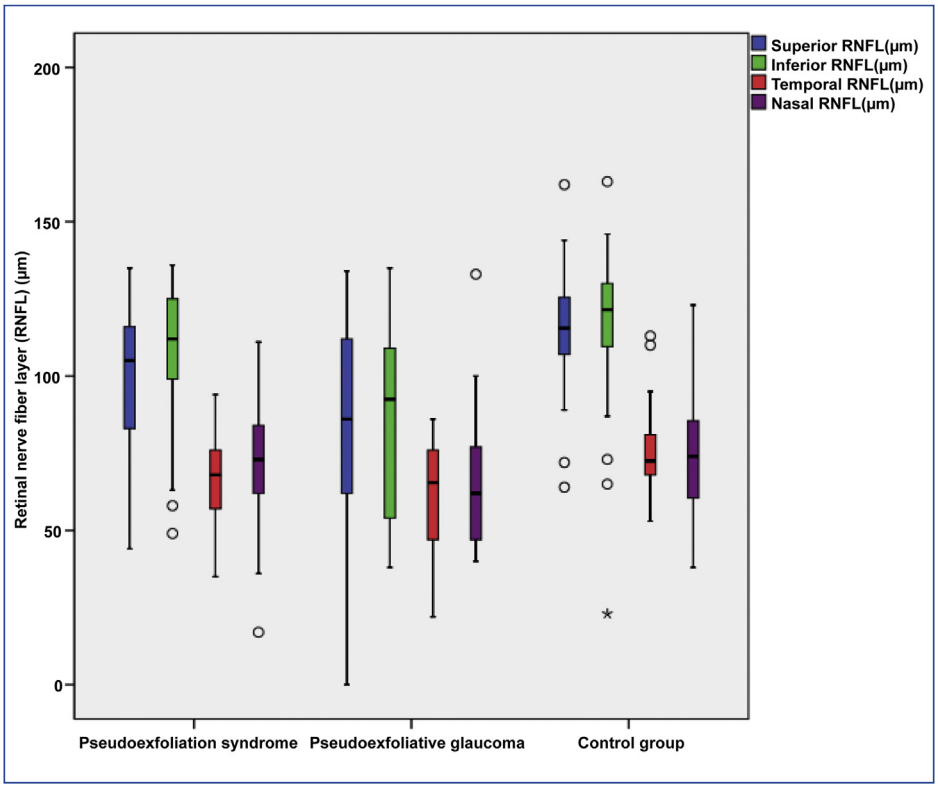

Figure 4. Retinal nerve fiber layer quadrant distribution by group.

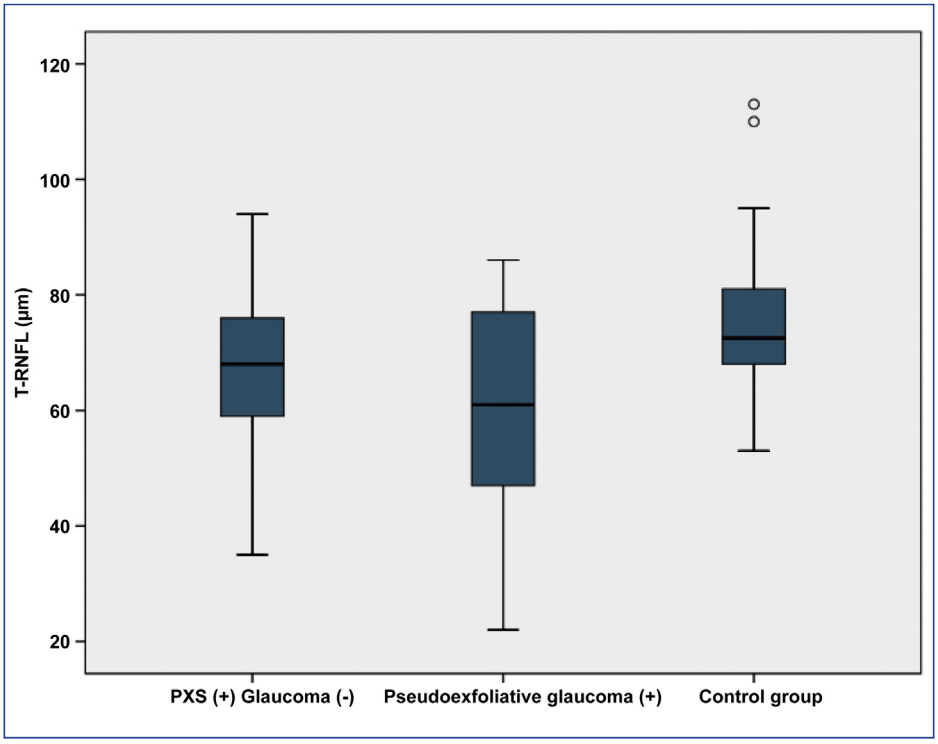

Figure 5. Temporal retinal nerve fiber layer quadrant distribution by group. T-RNFL: temporal retinal nerve fiber layer; PXS: pseudoexfoliation syndrome; PXG: pseudoexfoliative glaucoma.

Table 4 Relationship between total RNFL and PSV, EDV, and RI by group.

	Total RNFL PEX (+) Glaucoma (−) r	 *P*	PEX (+) Glaucoma (+) r	 *P*	Control r	 *P*
PSV	−0.164	0.240	−0.743	0.001**	0.317	0.036*
EDV	−0.207	0.138	−0.691	0.001**	0.337	0.025*
RI	0.095	0.499	−0.152	0.548	−0.046	0.766

RNFL: retinal nerve fiber layer; PEX: pseudoexfoliation; PSV: peak systolic velocity; EDV: end diastolic velocity; RI: resistive index.
* $P < 0.05$ (Pearson correlation analysis).
** $P < 0.01$ (Pearson correlation analysis).

retrobulbar circulation together with the decreased blood flow volume [19].

Harris et al. observed a correlation between OA circulation parameters and sex. They stated that aging may result in 30% and 12% reductions in OA EDV in women and men, respectively. They reported that aging was associated with an increase in the RI of the OA by 50% in women and 26% in men [17].

Martinez and Sanchez observed that hemodynamic parameters in retrobulbar vessels were significantly lower in patients with pseudoexfoliative glaucoma than those in patients with PXS. Additionally, in the same study, no difference was observed between patients with PXS and the control group regarding retrobulbar blood flow parameters [1].

In our study, PSV and EDV measurements of the OA were lower in patients with PEX with and without glaucoma than those in patients in the control group. On the other hand, no significant difference was observed between patients with PEX with and without glaucoma. Although no statistically significant difference was observed among the groups in terms of RI, the average RI was higher in groups with PEX.

Exfoliative material has been identified in the central retinal artery and short posterior ciliary arteries [3,20], which feed the prelaminar and laminar regions of the superficial nerve fiber layer. These vessels provide vascular support to the optic nerve head [1].

PXS is a systemic disease characterized by marked vascular damage [21,22]. Vascular changes occurring in eyes with PEX increase vascular permeability, resulting in occlusion and disappearance of vessels, neovascularization, and an increased RI of the OA. Pseudoexfoliative material accumulates in vascular endothelial cells, smooth muscle cells, and pericytes. All these cell types play regulatory roles in local microcirculation [7].

The severity of glaucoma in eyes with PEX is associated with the amount of pseudoexfoliative material deposited in the cribriform region of the trabecular meshwork [23]. In a 10-year study by Puska et al., 38% of patients with unilateral PXS progressed to bilateral PXS [24]. Therefore, follow-up of patients with PXS in one eye is important.

Many studies have revealed that the RNFL in the inferotemporal quadrant was significantly thinner in eyes with PXS than that in the control group. This finding may be an early sign of retinal nerve fiber damage in eyes with PXS, despite a normal IOP and intact visual field [13]. Puska et al. suggested that pseudoexfoliative material may be a risk factor independent of IOP for ganglion cell damage in eyes with PXS and glaucoma [24].

In this study, the total RNFL was thicker in eyes of patients in the control group than that in eyes of patients with PEX without glaucoma. Similarly, the total RNFL was thicker in eyes of patients with PEX without glaucoma than that in eyes of patients with PEX and glaucoma.

The RNFL S, RNFL I, and RNFL T were thicker in the control group than in the other groups. However, no significant difference was noted among the groups regarding RNFL N thickness. Furthermore, when patients with PEX with and without glaucoma were compared, no significant difference was observed except for that in RNFL S thickness, which was lower in patients with glaucoma. Changes in the optic nerve head structure and RNFL thickness may precede the development of visual field loss in eyes of patients with glaucoma. Therefore, segmental reduction of the RNFL in eyes with PEX may be an early sign of glaucomatous damage [11]. In our study, the RNFL values of the patients in Groups A and B were similar, possibly because the decrease in RNFL thickness occurred before visual field loss.

Study limitations

This study has several limitations. First, our sample was small, limiting the generalizability of the results. Second, the follow-up period for patients with PEX in only one eye was too short to monitor the condition of the other eye. PEX may subsequently develop in the other eye, and the OA blood flow parameters may change. Third, we did not apply the visual field test to all cases included in the study; however, visual field testing was performed in cases with glaucoma or suspected glaucoma. Finally, parameter changes that may have been due to globe compression could not be considered during CDI.

Conclusion

Our study showed a decrease in total RNFL thickness in eyes with PEX. PXS with or without glaucoma was associated with a decrease in the PSV and EDV of the OA. An extensive study may be needed to further explore the role of PXS in OA blood flow parameters.

Ethical approval

This prospective study was conducted at the Adatip Hospital in accordance with the principles of the Declaration of Helsinki. The study was reviewed and approved by the Private Istanbul Medipol University Ethics Committee (10840098-604.01.01-E.12134/10.03.2020).

Acknowledgments

None.

Disclosure of interest

The authors declare that they have no competing interest.

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